

SEQUENCE LISTING

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TECH CENTER 1600/2000

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<120> MUTANT KANAMYCIN NUCLEOTIDYLTRANSFERASE AND A METHOD OF SCREENING THERMOPHILIC BACTERIA USING THE SAME

<130> 04853.0048-00000

<140> 09/697,186

<141> 2000-10-27

<150> JP 309616/1999

<151> 1999-10-29

<160> 20

<170> PatentIn Ver. 2.1

<210> 1

<211> 253

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Mutant enzyme
 obtained by introduction of point mutation into
 wild type KNT gene of Staphylococcus aureus and
 its expression /

<400> 1

Met Asn Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val 1 5 10

His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr 35 40 45

Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Glu Ala Glu Phe 50 55 60

Ser His Glu Trp Thr Thr Gly Glu Trp Lys Val Glu Val Asn Phe Tyr 65 70 75 80

Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Gln Val Glu Ser Asp Trp 85 90 95

Pro Leu Thr His Gly Gln Phe Phe Ser Ile Leu Pro Ile Tyr Asp Ser 100 105 110 Gly Gly Tyr Leu Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala 115 120 125

Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe 130 135 140

Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr 145 150 155 160

Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu 165 170 175

Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Ser Val Leu 180 185 190

Thr Glu Ala Val Lys Gln Ser Asp Leu Pro Ser Gly Tyr Asp His Leu 195 200 205

Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu 210 215 220

Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Thr Glu Arg 235 230 240

His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe 245 250

<210> 2

<211> 253

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Mutant enzyme obtained by introduction of point mutation into wild type KNT gene of Staphylococcus aureus and its expression

<400> 2

Met Lys Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val 1 5 10 15

His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys 20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr 35 40 45

Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Gly Ala Glu Phe 50 55 60

Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr 65 70 75 80

Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Arg Val Glu Ser Asp Trp 85 90 95 Pro Leu Thr His Gly Arg Phe Phe Ser Ile Leu Pro Ile Tyr Asp Pro 100 105 110

Gly Gly Tyr Phe Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala 115 120 125

Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe 130 135 140

Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr 145 150 155 160

Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu 165 170 175

Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Ser Val Leu 180 185 190

Thr Glu Ala Val Lys Gln Pro Asp Leu Pro Ser Gly Tyr Asp His Leu 195 200 205

Cys Gln Leu Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu 210 215 220

Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Thr Glu Arg 225 230 235 240

His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe 245 250

<210> 3

<211> 253

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Mutant enzyme obtained by introduction of point mutation into wild type KNT gene of Staphylococcus aureus and its expression

<400> 3

Met Lys Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val 1 5 10 15

His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr 35 40 45

Ser Asp Ile Glu Met Met Cys Val Leu Ser Thr Glu Gly Val Glu Phe 50 60

Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr 65 70 75 80

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										4							
	Ser	Glu	Glu	Ile	Leu 85	Leu	Asp	Tyr	Ala	Ser	Arg	Val	Glu	Pro	Asp 95	Trp	
	Pro	Leu	Thr	His 100	Gly	Arg	Phe	Phe	Ser 105	Ile	Leu	Pro	Ile	Tyr 110	Asp	Pro	
	Gly	Gly	Tyr 115	Phe	Glu	Lys	Val	Tyr 120	Gln	Thr	Ala	Lys	Ser 125	Val	Glu	Ala	
	Gln	Lys 130	Phe	His	Asp	Ala	Ile 135	Cys	Ala	Leu	Ile	Val 140	Glu	Glu	Leu	Phe	
	Glu 145	_	Ala	Gly	Lys	Trp 150	Arg	Asn	Ile	Arg	Val 155	Gln	Gly	Pro	Thr	Thr 160	
	Phe	Leu	Pro	Ser	Leu 165	Thr	Val	Gln	Val	Ala 170	Met	Ala	Gly	Ala	Met 175	Leu	
	Ile	Gly	Leu	His 180	His	Arg	Ile	Cys	Tyr 185	Thr	Thr	Ser	Ala	Ser 190	Val	Leu	
	Thr	Glu	Al a 195	Val	Lys	Gln	Pro	Asp 200	Leu	Pro	Pro	Gly	Tyr 205	Val	Gln	Leu	
	Cys	Gln 210	Leu	Val	Met	Ser	Gly 215	Gln	Leu	Ser	Asp	Pro 220	Glu	Lys	Leu	Leu	
	Glu 225		Leu	Glu	Asn	Phe 230	Trp	Asn	Gly	Val	Gln 235	Glu	Trp	Ala	Glu	Arg 240	
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		0> 4		-	ccgtt		eggeg	ggata	a tgg	jta							3!
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<210> 6 <211> 35 <212> DNA <213> Artificial Sequence	
<220> <223> Description of Artificial Sequence: Primer for PCR amplification	
<400> 6 gactgtacgg aattcgagct cgagcaaatc taaaa	35
<210> 7 <211> 35 <212> DNA <213> Artificial Sequence	
<220> <223> Description of Artificial Sequence: Primer for subcloning of WT	
<400> 7 gactgtacgc atatgaatgg accaataata atgac	35
<pre><210> 8 <211> 35 <212> DNA <213> Artificial Sequence</pre>	
<220> <223> Description of Artificial Sequence: Primer for subcloning of KT3-11 and HTK	
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<210> 9 <211> 35 <212> DNA <213> Artificial Sequence	
<220> <223> Description of Artificial Sequence: Primer for subcloning	
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<210> 10 <211> 759 <212> DNA	

<213> Staphylococcus aureus

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_	_				ttc Phe 230					_					_
				_	gat Asp	-				_		_			
<210> 11 <211> 253 <212> PRT <213> Staphylococcus aureus															
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His	Glu	Ile	-		Arg			_	Lys	_	_	_	_		Lys
Ala	Ile	Gly 35	Val	Tyr	Gly	Ser	Leu 40	Gly	Arg	Gln	Thr	Asp 45	Gly	Pro	Tyr
Ser	Asp 50	Ile	Glu	Met	Met	Cys 55	Val	Met	Ser	Thr	Glu 60	Glu	Ala	Glu	Phe
Ser 65	His	Glu	Trp	Thr	Thr 70	Gly	Glu	Trp	Lys	Val 75	Glu	Val	Asn	Phe	Asp 80
Ser	Glu	Glu	Ile	Leu 85	Leu	Asp	Tyr	Ala	Ser 90	Gln	Val	Glu	Ser	Asp 95	Trp
Pro	Leu	Thr	His 100	Gly	Gln	Phe	Phe	Ser 105	Ile	Leu	Pro	Ile	Tyr 110	Asp	Ser
Gly	Gly	Tyr 115	Leu	Glu	Lys	Val	Tyr 120	Gln	Thr	Ala	Lys	Ser 125	Val	Glu	Ala
Gln	Thr 130	Phe	His	Asp	Ala	Ile 135	Cys	Ala	Leu	Ile	Val 140	Glu	Glu	Leu	Phe
Glu 145	Tyr	Ala	Gly	Lys	_	_		Ile	_			Gly	Pro	Thr	Thr 160
Phe	Leu	Pro	Ser	Leu 165	Thr	Val	Gln	Val	Ala 170	Met	Ala	Gly	Ala	Met 175	Leu
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Thr	Glu	Ala 195	Val	Lys	Gln	Ser	Asp 200	Leu	Pro	Ser	Gly	Tyr 205	Asp	His	Leu

Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu 210 215 220

Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Thr Glu Arg 225 230 235 240

His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe 245 250

<210> 12

<211> 253

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Mutant enzyme obtained by introduction of point mutation into wild type KNT gene of Staphylococcus aureus and its expression

<400> 12

Met Asn Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val 1 5 10 15

Tyr Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr 35 40 45

Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Glu Ala Glu Phe 50 55 60

Ser His Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr 65 70 75 80

Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Arg Val Glu Ser Asp Trp
85 90 95

Pro Leu Thr His Gly Arg Phe Phe Ser Ile Leu Pro Ile Tyr Asp Pro 100 105 110

Gly Gly Tyr Phe Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala 115 120 125

Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe 130 135 140

Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr 145 150 155 160

Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu 165 170 175

Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Ser Val Leu 180 185 190 Thr Glu Ala Leu Lys Gln Ser Asp Leu Pro Ser Gly Tyr Asp His Leu 195 200 205

Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu 210 220

Glu Ser Leu Glu Asn Phe Trp Asn Gly Val Gln Glu Trp Ala Glu Arg 225 230 235 240

His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe 245 250

<210> 13

<211> 253

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Mutant enzyme obtained by introduction of point mutation into wild type KNT gene of Staphylococcus aureus and its expression

<400> 13

Met Asn Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val 1 5 10 15

His Glu Ile Lys Glu Arg Ile Leu Asn Lys Tyr Gly Asp Asp Val Lys
20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr 35 40 45

Ser Asp Ile Glu Met Met Cys Val Leu Ser Thr Glu Gly Val Glu Phe 50 55 60

Ser His Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr 65 70 75 80

Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Gln Val Glu Pro Asp Trp
85 90 95

Pro Leu Thr His Gly Arg Phe Phe Ser Ile Leu Pro Ile Tyr Asp Ser 100 105 110

Gly Gly Tyr Leu Gly Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala 115 120 125

Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe 130 135 140

Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr 145 150 155 160

Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu 165 170 175 Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Leu Val Leu 180 185 190

Thr Glu Ala Val Lys Gln Ser Asp Leu Pro Ser Gly Tyr Asp His Leu 195 200 205

Cys Gln Leu Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu 210 215 220

Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Thr Glu Arg 225 230 235 240

His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe 245 250

<210> 14

<211> 253

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Mutant enzyme obtained by introduction of point mutation into wild type KNT gene of Staphylococcus aureus and its expression

<400> 14

Met Asn Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val

His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys 20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr 35 40 45

Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Gly Ala Glu Phe 50 60

Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr 65 70 75 80

Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Arg Val Glu Pro Asp Trp 85 90 95

Pro Leu Thr His Gly Arg Phe Phe Ser Ile Leu Pro Ile Tyr Asp Ser 100 105 110

Gly Gly Tyr Leu Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala 115 120 125

Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe 130 135 140

Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr 145 150 150 160 Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu 165 170 175

Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Ser Val Leu 180 185 190

Thr Glu Ala Val Lys Gln Pro Asp Leu Pro Ser Gly Tyr Asp His Leu 195 200 205

Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu 210 215 220

Glu Ser Leu Glu Asn Phe Trp Asn Gly Val Gln Glu Trp Thr Glu Arg 235 240

His Gly Tyr Ile Val Asn Val Ser Lys Arg Ile Pro Phe 245 250

<210> 15

<211> 253

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Mutant enzyme obtained by introduction of point mutation into wild type KNT gene of Staphylococcus aureus and its expression

<400> 15

Met Ser Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val

His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr 35 40 45

Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Gly Ala Glu Phe 50 55 60

Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr 65 75 80

Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Arg Val Glu Ser Asp Trp 85 90 95

Pro Leu Thr His Gly Arg Phe Phe Ser Ile Leu Pro Ile Tyr Asp Pro 100 105 110

Gly Gly Tyr Phe Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala 115 120 125

Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe 130 135 140 Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr 145 150 155 160

Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu 165 170 175

Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Thr Val Leu 180 185 190

Thr Glu Ala Val Lys Leu Ser Asp Leu Pro Ser Gly Tyr Asp His Leu 195 200 205

Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu 210 215 220

Glu Ser Leu Glu Asn Phe Trp Asn Gly Val Gln Glu Trp Thr Glu Arg 225 230 235 240

His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe 245 250

<210> 16

<211> 253

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Mutant enzyme obtained by introduction of point mutation into wild type KNT gene of Staphylococcus aureus and its expression

<400> 16

Met Asn Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val 1 5 10 15

His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr 35 40 45

Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Glu Thr Glu Phe 50 55 60

Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr 65 70 75 80

Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Arg Val Glu Pro Asp Trp 85 90 95

Pro Leu Thr His Gly Lys Phe Phe Ser Ile Leu Pro Ile Tyr Asp Thr 100 105 110

Gly Gly Tyr Leu Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala 115 120 125 Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe 130 135 140

Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Leu Thr 145 150 155 160

Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu 165 170 175

Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Gly Ala Ser Val Leu 180 185 190

Thr Glu Ala Val Arg Gln Pro Asp Leu Pro Pro Gly Tyr Asp His Leu 195 200 205

Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu 210 215 220

Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Ala Glu Arg 225 230 235 240

His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe 245 250

<210> 17

<211> 253

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Mutant enzyme obtained by introduction of point mutation into wild type KNT gene of Staphylococcus aureus and its expression

<400> 17

Met Asn Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val 1 5 10 15

His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr 35 40 45

Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Gly Ala Glu Phe 50 55 60

Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr 65 70 75 80

Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Arg Val Glu Ser Asp Trp 85 90 95

Pro Leu Thr His Gly Arg Phe Phe Ser Ile Leu Pro Ile Tyr Asp Pro

Gly Gly Tyr Phe Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala 115 120 125

Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe 130 135 140

Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr 145 150 155 160

Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu 165 170 175

Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Ser Val Leu 180 185 190

Thr Glu Ala Val Lys Gln Pro Asp Leu Pro Ser Gly Tyr Val Gln Leu 195 200 205

Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu 210 215 220

Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Thr Glu Arg 225 230 235 240

His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe 245 250

<210> 18

<211> 253

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Mutant enzyme obtained by introduction of point mutation into wild type KNT gene of Staphylococcus aureus and its expression

<400> 18

Met Asn Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val 1 5 10 15

His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr 35 40 45

Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Gly Ala Glu Phe 50 55 60

Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr 65 70 75 80

Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Arg Val Glu Ser Asp Trp 85 90 95 Pro Leu Thr His Gly Arg Phe Phe Ser Ile Leu Pro Ile Tyr Asp Pro 100 105 110

Gly Gly Tyr Phe Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala 115 120 125

Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe 130 135 140

Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr 145 150 155 160

Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu 165 170 175

Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Ser Val Leu 180 185 190

Thr Glu Ala Val Lys Gln Pro Asp Leu Pro Ser Gly Tyr Val Gln Leu 195 200 205

Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu 210 215 220

Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Thr Glu Arg 235 230 235 240

His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe 245 250

<210> 19

<211> 253

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Mutant enzyme obtained by introduction of point mutation into wild type KNT gene of Staphylococcus aureus and its expression

<400> 19

Met Asn Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val 1 5 10 15

His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr 35 40 45

Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Gly Ala Glu Phe 50 55 60

Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr 65 70 75 80

Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Arg Val Glu Pro Asp Trp
85 90 95

Pro Leu Thr His Gly Lys Phe Phe Ser Ile Leu Pro Ile Tyr Asp Pro 100 105 110

Gly Gly Tyr Leu Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala 115 120 125

Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe 130 135 140

Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr 145 150 155 160

Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu 165 170 175

Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Ser Val Leu 180 185 190

Thr Glu Ala Val Lys Gln Pro Asp Leu Pro Ser Gly Tyr Val Gln Leu 195 200 205

Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Pro Glu Lys Leu Leu 210 215 220

Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Thr Glu Arg 225 230 235 240

His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe 245 250

<210> 20

<211> 253

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Mutant enzyme obtained by introduction of point mutation into wild type KNT gene of Staphylococcus aureus and its expression

<400> 20

Met Asn Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val 1 5 10 15

His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr 35 40 45

Ser Asp Ile Glu Met Met Cys Val Leu Ser Thr Glu Glu Ala Glu Phe 50 55 60

- Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr 65 70 75 80
- Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Arg Val Glu Pro Asp Trp 85 90 95
- Pro Leu Thr His Gly Lys Phe Phe Ser Ile Leu Pro Ile Tyr Asp Pro 100 105 110
- Gly Gly Tyr Leu Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala 115 120 125
- Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe 130 135 140
- Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr
 145 150 155 160
- Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu 165 170 175
- Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Ser Val Leu 180 185 190
- Thr Glu Ala Val Lys Gln Pro Asp Leu Pro Ser Gly Tyr Val Gln Leu 195 200 205
- Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu 210 220
- Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Thr Glu Arg 225 230 235 240
- His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe 245 250